

Research Brief: Rapid Screens for Chlorinated Hydrocarbons (Dioxins) in Contaminated Sediments

Issue

Dioxins (polychlorinated dibenzo-p-dioxins and diobenzofurans) are persistent and widespread contaminants of the sediments of many industrialized waterways requiring maintenance dredging. When dioxins are suspected of being present in channel sediments during permit evaluation prior to dredging, chemical analyses are usually required. The cost of high resolution GC/MS analysis of a single sediment sample can be as much as \$2,000. When a large number of samples have to be analyzed, the costs can be a burden to the applicant. Low-cost screening assays allow early identification of sediments that are demonstrably contaminated, thus eliminating them from further costly chemical analysis. Screening assays for dioxins have other applications as well, such as monitoring the progress and effectiveness of remediation efforts.

Research/Objectives

The objective of this work is to evaluate, refine, and validate the use of cultured vertebrate cell bioassays coupled with rapid extraction and sample cleanup techniques For application as screening tests for dioxins. The first steps in the toxin mechanism of action of dioxin and related compounds involve binding of the chemicals with a soluble receptor in the cytoplasm of the cells of a vertebrate organism. The receptor/ligand complex is then translocated to the nucleus of the cell where it attaches to recognition sites on the DNA of the cell and initiates processes that result in the synthesis of gene products such as enzymes. Bioassays using cultured cells of mammals and fish can be used to detect and quantitate the dioxin-like activity in extracts of environmental samples by taking advantage of this mechanism. Selected gene products resulting from exposure of the cells to the extracts can be measured quantitatively and related to the presence of dioxin-like chemicals. By coupling cell-based bioassay methods with accelerated solvent extraction (ASE), which uses high pressure and temperature to extract chemicals from sediments and soils, a rapid, low-cost screening assay for dioxins can be produced. Research will be conducted on several cell lines, comparing them with each other and with chemical analyses of sediments. An optimized method for a cell-based assay will be field-demonstrated.

Results/Products

Cell-based assays detect and measure the activity of dioxin and dioxin-like chemicals by employing the same cellular machinery responsible for the toxicity of these chemicals in whole organisms. Advantages are cost and time savings. For example, the difference in cost for GC/MS analysis of dioxins is at least one order of magnitude, but may be more, depending on the number of samples per batch, the number of sample replicates, and other factors. For these reasons, and because the sensitivity of the cell-

based assays are potentially of the same order as GC/MS, the cells can provide screening to prioritize sites for definitive chemical analysis when such analyses are required for regulatory or other purposes.

Results/Products (Cont.)

When cleanup and remediation involving soils or sediments contaminated with PAH, PCB, or dioxin are issues, the cells also offer a less expensive alternative to chemistry for monitoring the effectiveness of treatments. Lower costs allow more samples to be assayed, resulting in better characterization of the extent of contamination at a site. To date, H4IIE rat hepatoma and recombinant human 101L cell (P450 Reporter Gene System) assays have been formatted for 96-well plates and compliant instrumentation, and have been combined with a one-step ASE cleanup/extraction method. Detailed protocols have been written and methods have been compared. A field demonstration is in progress.

Technical Note DOER-C1 - Guidance for Performance of H4IIE Dioxin Screening Assay
Technical Note DOER-C8 - Comparison of Dioxin Screening Assays

Research Team

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